

# **Zebrafish Spawning and Reproduction From a Researcher's Perspective**

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**Daniel Castranova**

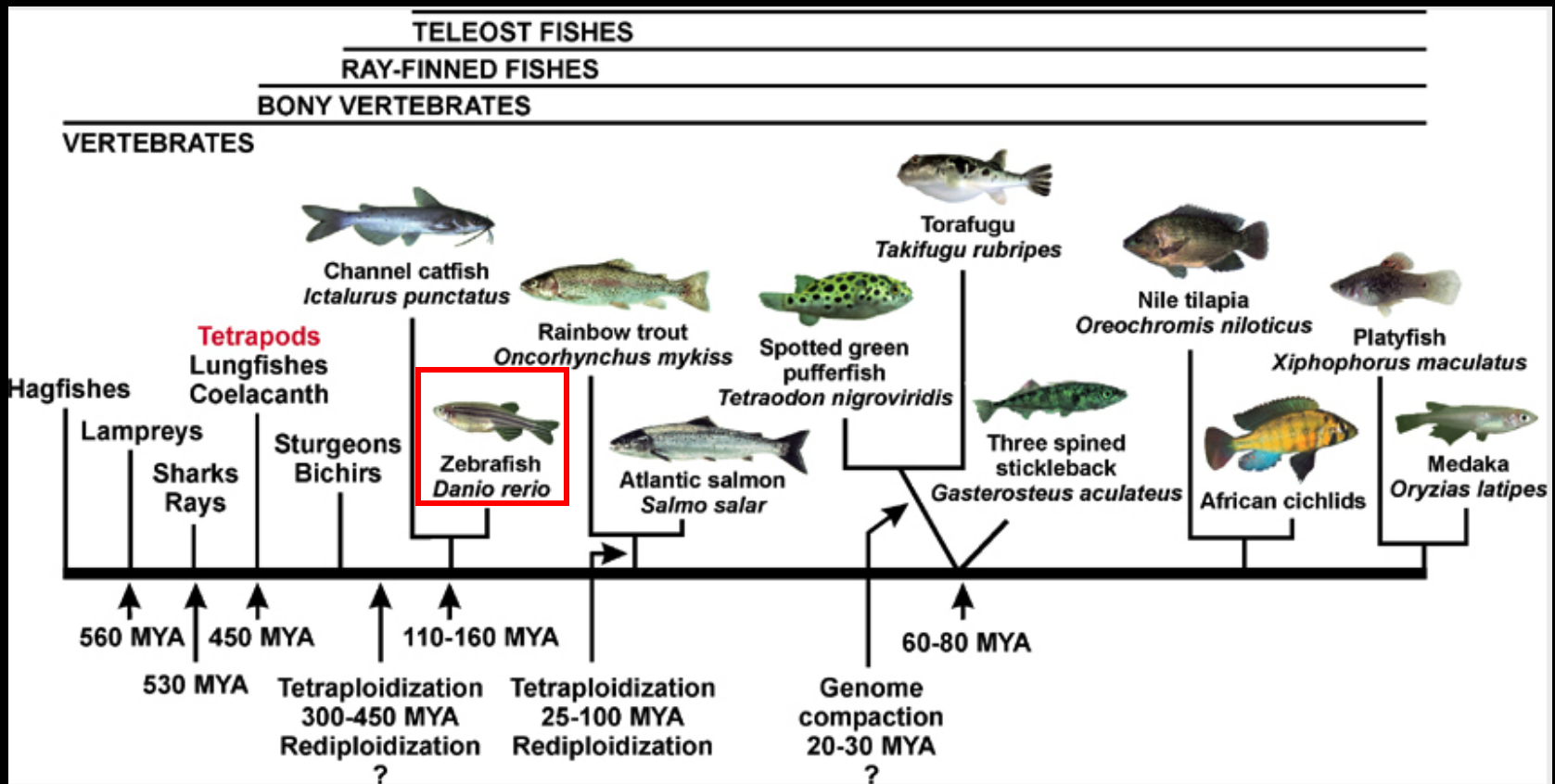
Aquatic Specialist (Charles River)

SVO, PGD, NICHD, NIH

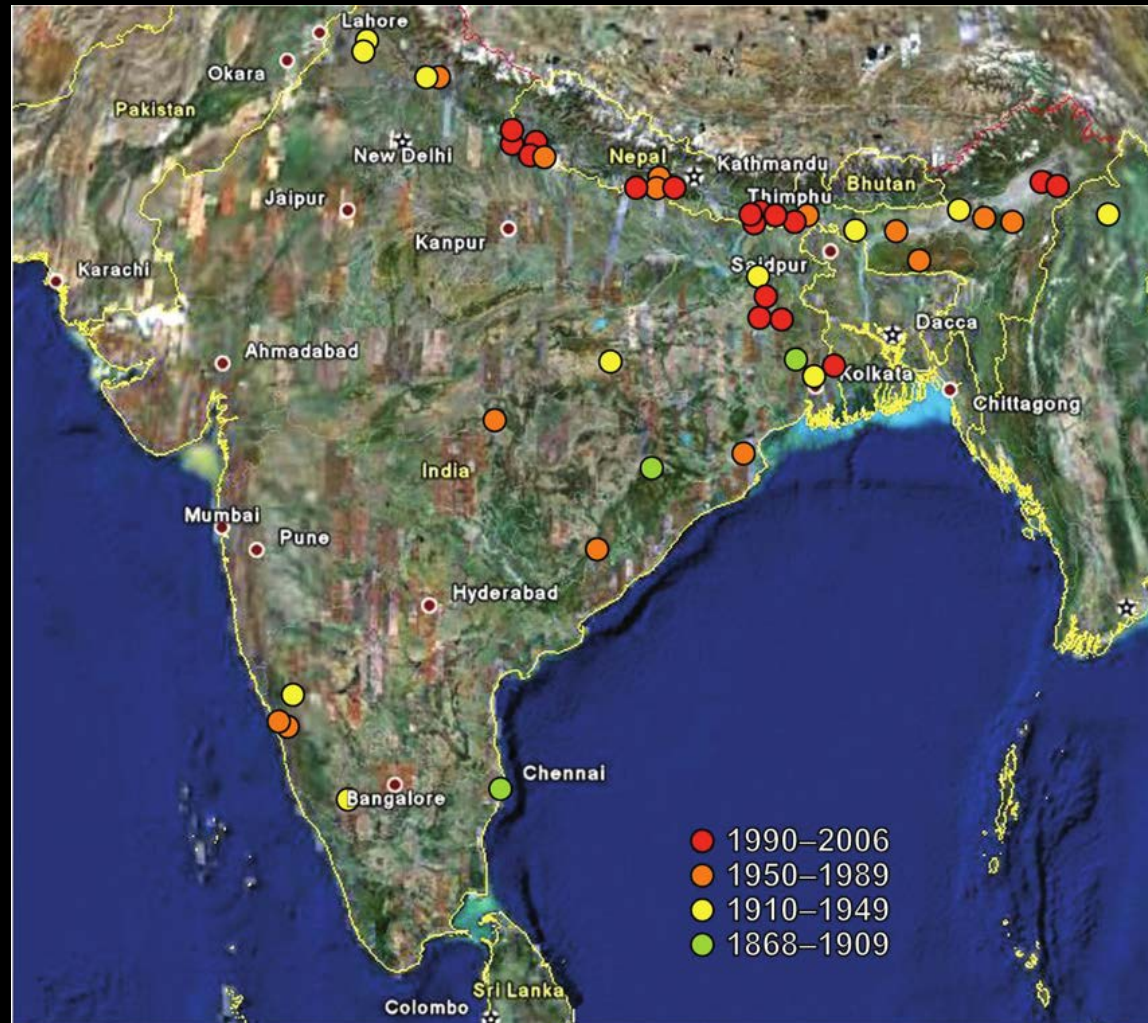
[castrand@mail.nih.gov](mailto:castrand@mail.nih.gov)

# The Zebrafish

- Danio rerio*



# Where do they come from?



Zebrafish in the Wild: a Review of Natural History and New Notes from the Field. R. E. Engeszer et al. 2007, Zebrafish. 4(1) 21-40.

Zebraphish in the Wild: a Review of Natural  
History and New Notes from the Field.  
R. E. Engeszer et al. 2007, Zebraphish.  
4(1) 21-40.

# Zebrafish in the Wild

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- Prefer shallow stagnant waters close to rivers and streams
- Seasonal spawners (summer)
- Shoaling fish
- Omnivores
- Tolerate a wide range of water quality parameters

# Zebrfish in the Lab

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- Mainly used as broodstock
- Sexually mature around 3 months
- Spawn well from 3 months to 2-2.5 years
- Breed all year round
- Many different breeding techniques

# Breeding

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- Off of System

Movie by Mark A. McElwain

# **Separating Males and Females**

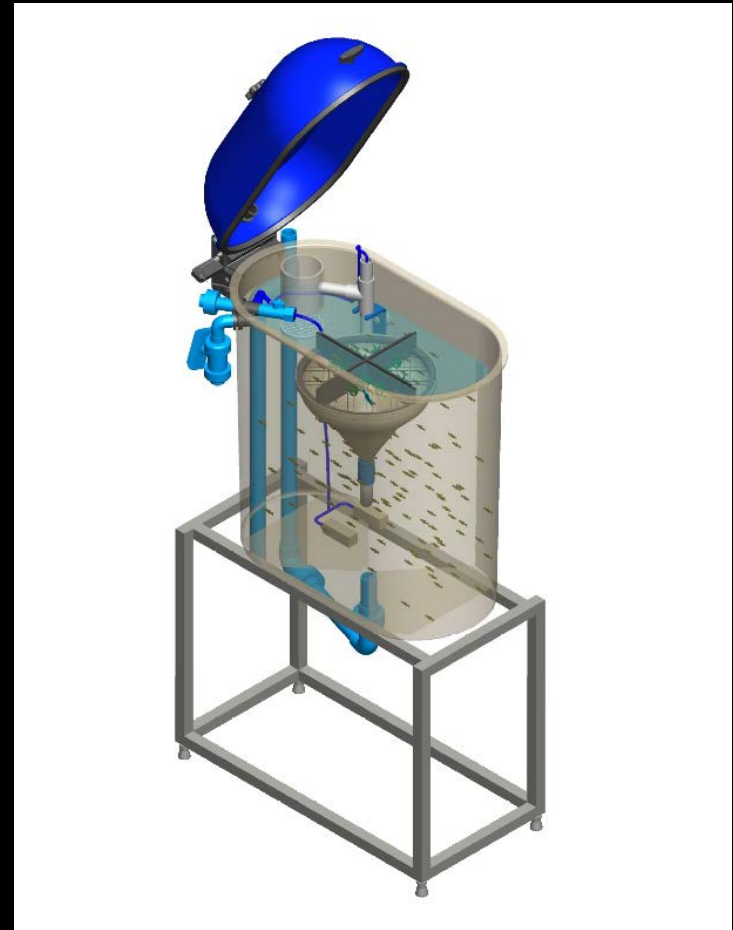
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# Large Scale Breeding

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- On System
    - MEPS (Mass Embryo Production System)
- By Aquatic Habitats



# Large Scale Breeding

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- Off System

Designed by Issac Adato  
and Christian Lawrence,  
Distributed by  
Techniplast



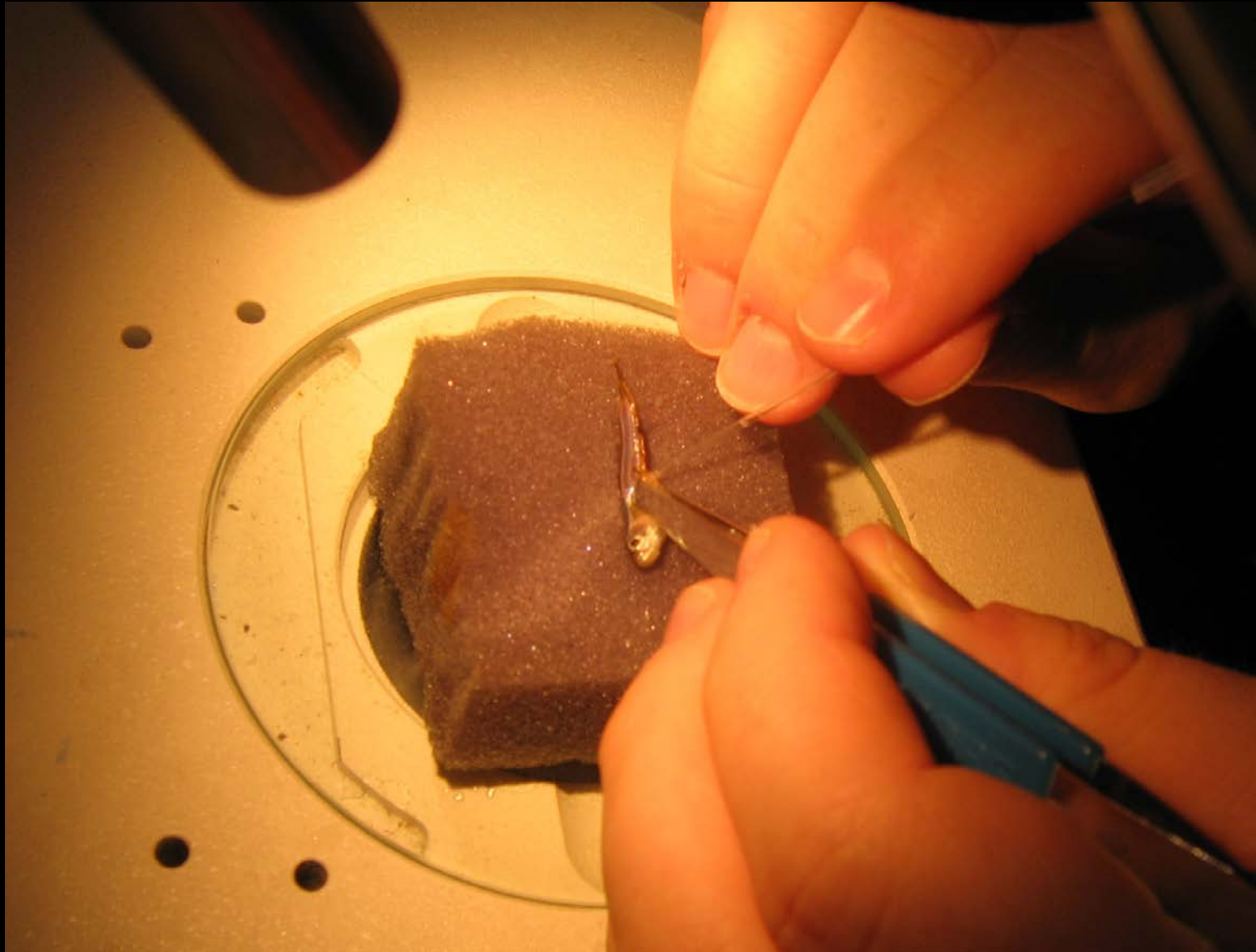
# If they won't Cooperate...

## In Vitro Fertilization

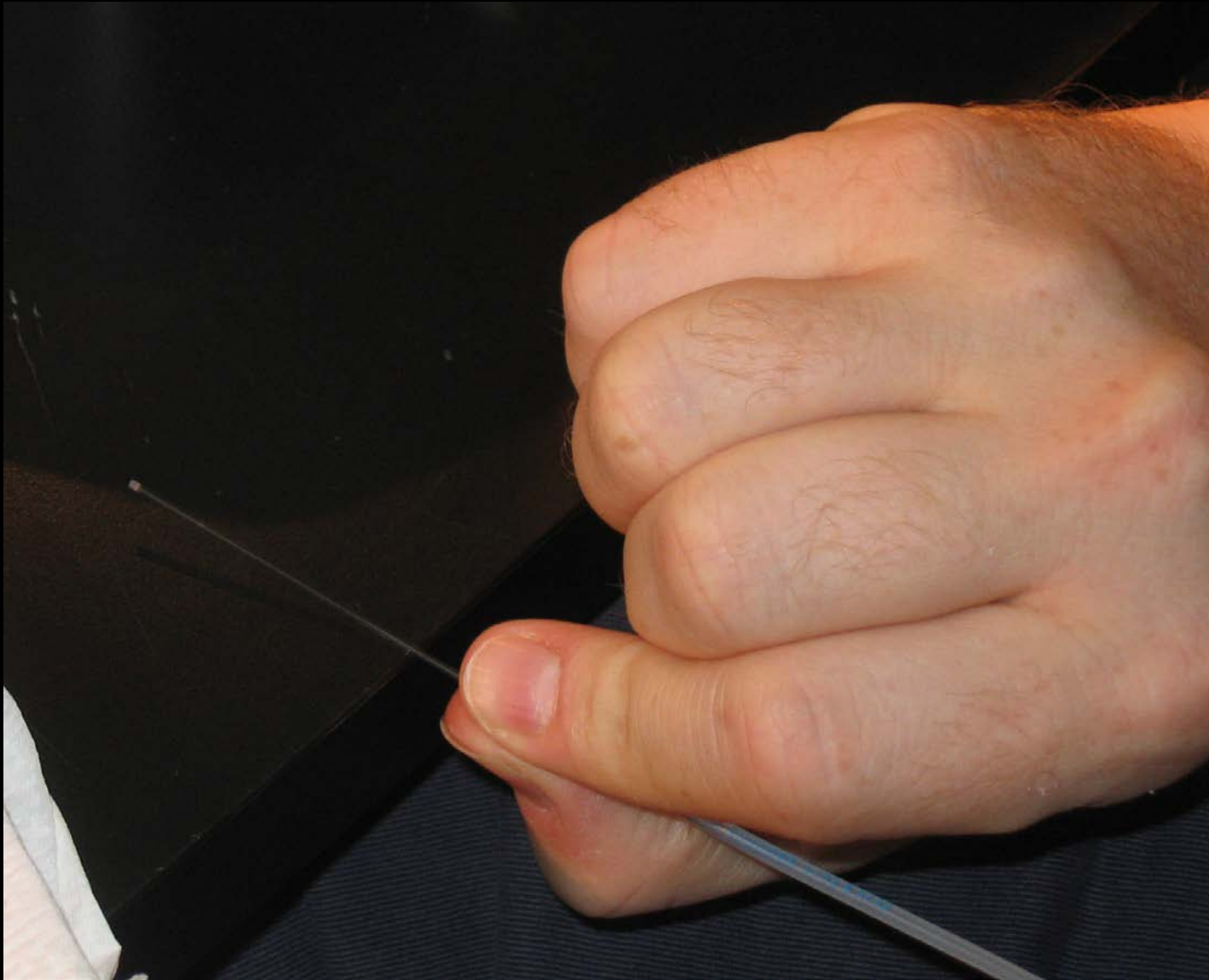
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- Older Animals
- “Compromised” Animals
- Synchronized Embryos
- Quarantine X Non-Quarantine
- ~ 10% Mortality Rate

# Mouth Pipette Sperm from the Male



# Sperm Sample

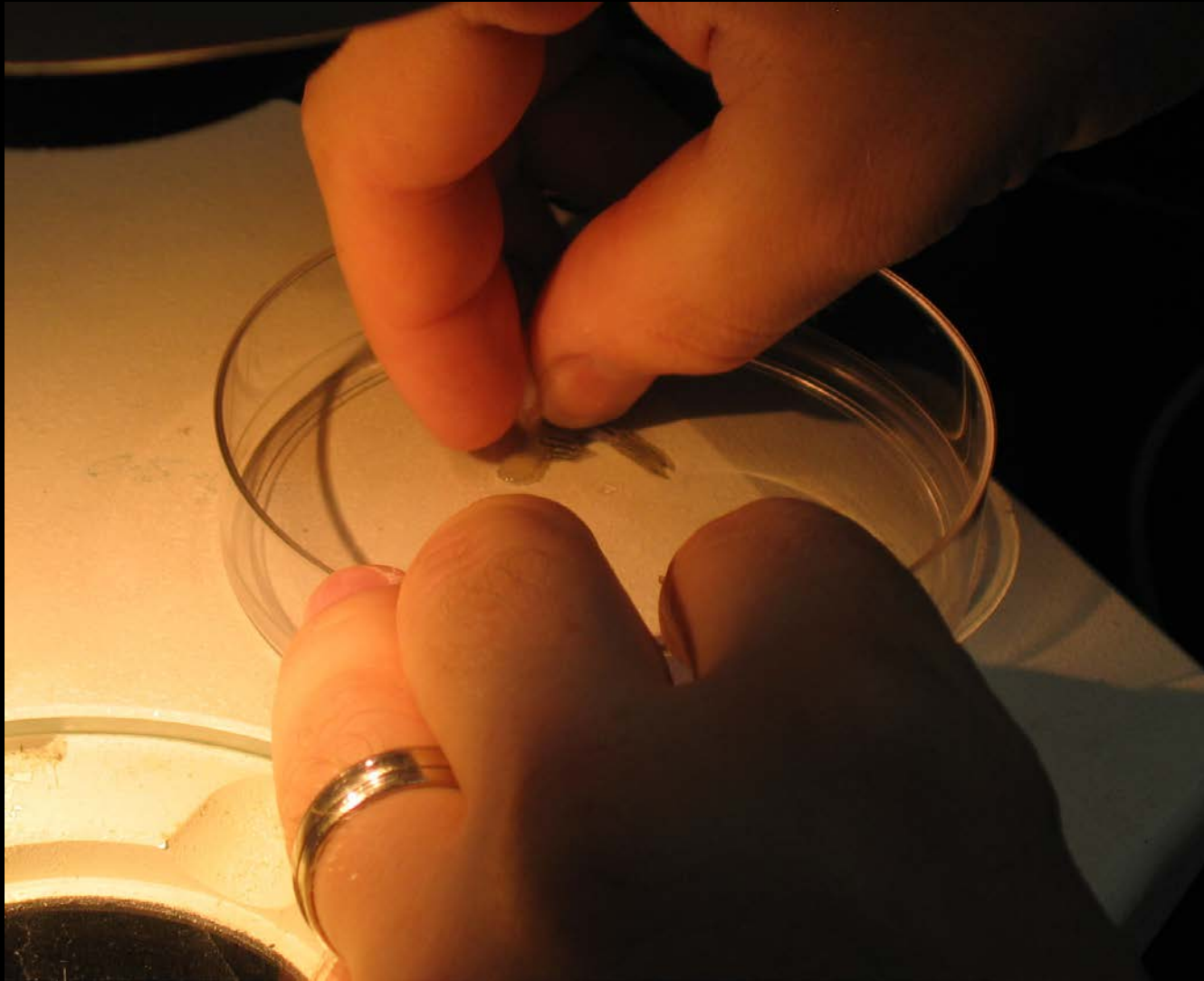




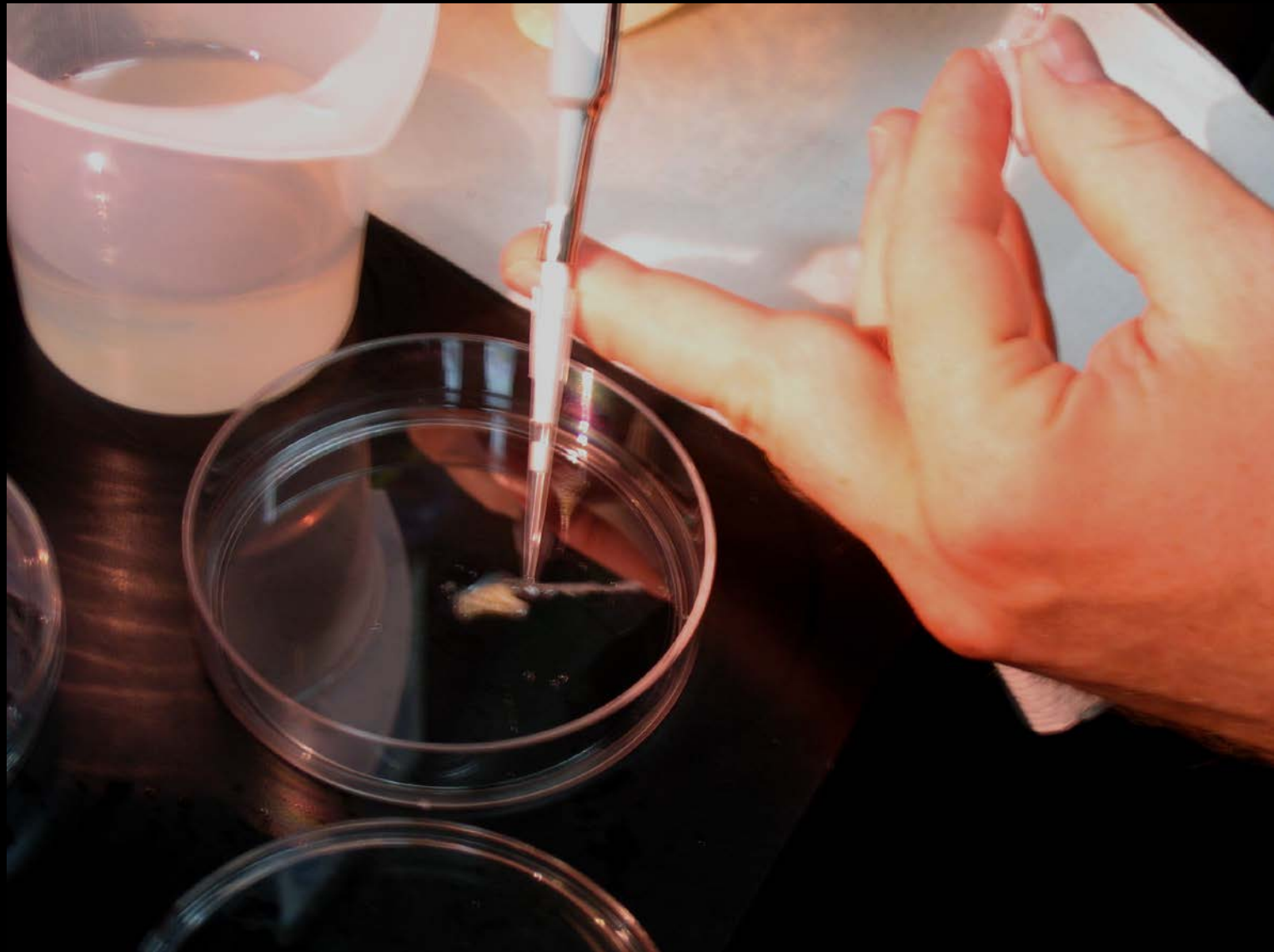
# Dilute Sperm in Hank's Salt Solution



# Squeeze the Females



# Sperm on Egg





# Just Add Water

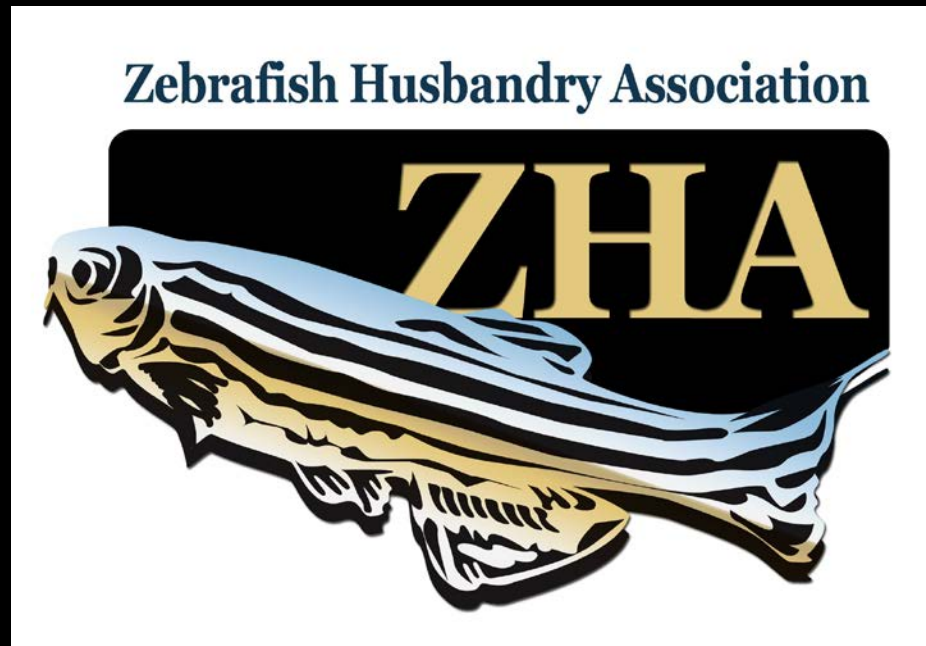


# The next 17 hours...

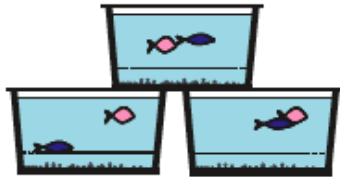
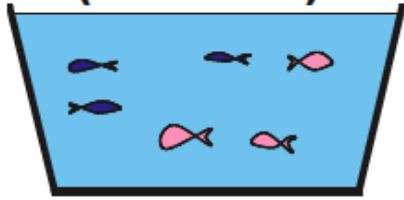
Rolf Karlstrom and Don Kane; Development 123:461, 1996

# Improving Reproduction and Spawning

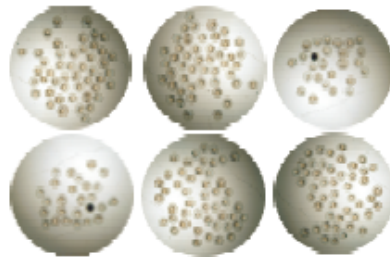
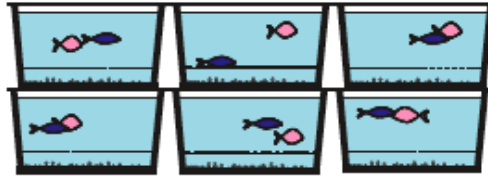
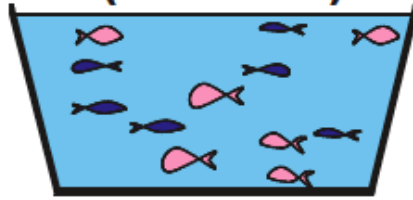
Insight from a collaborative research project organized through ZHA



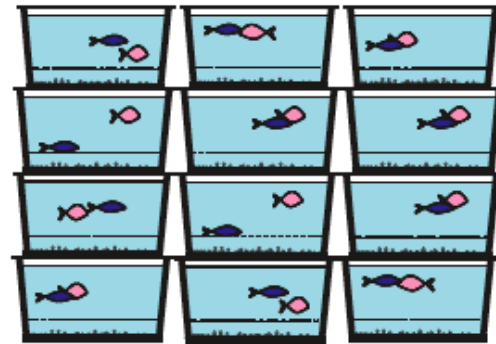
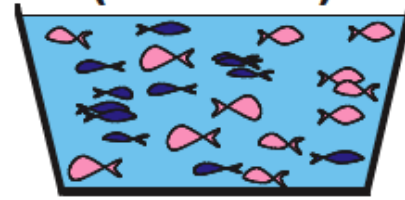
**3 fish per liter  
(4 tanks)**



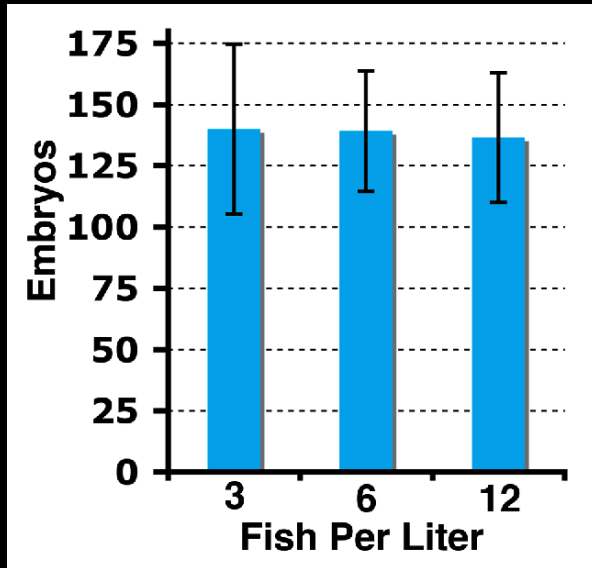
**6 fish per liter  
(2 tanks)**



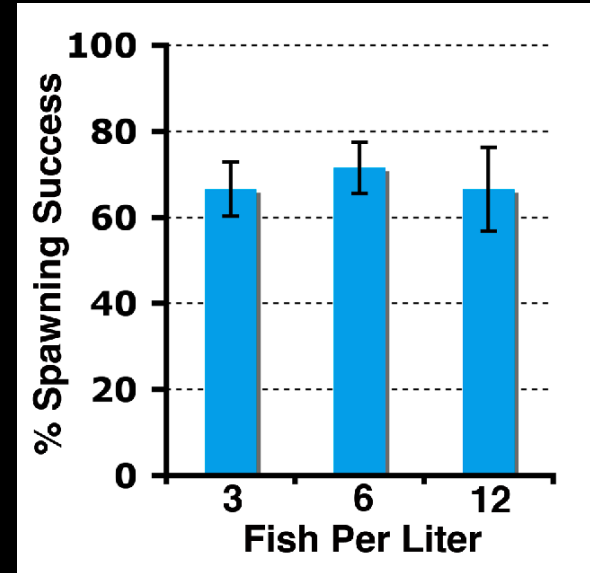
**12 fish per liter  
(2 tanks)**



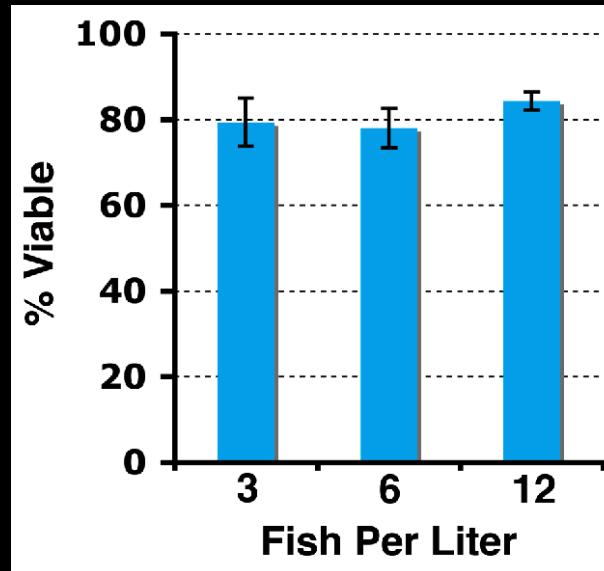
## Average Clutch Size



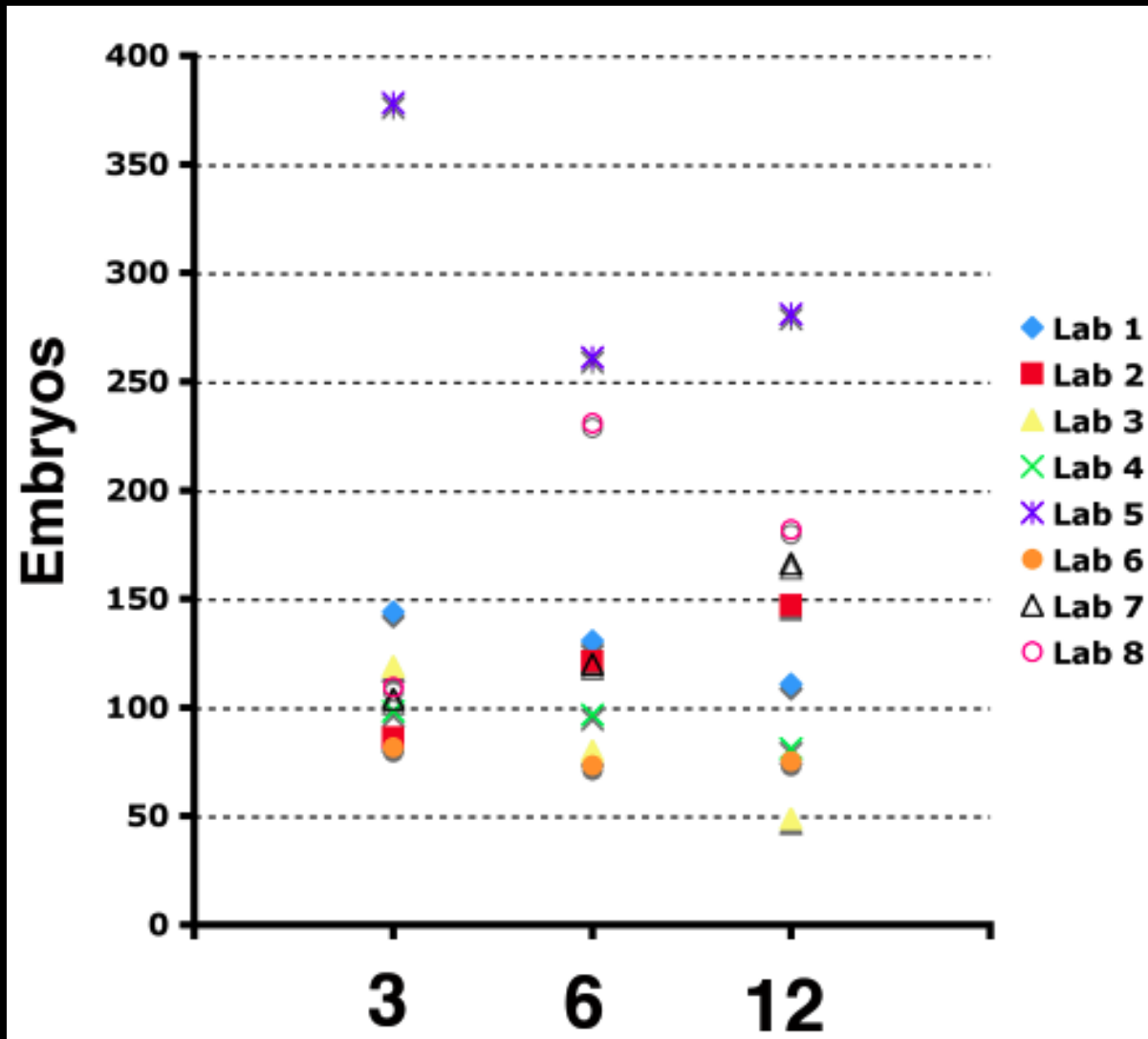
## Percent Spawning Success



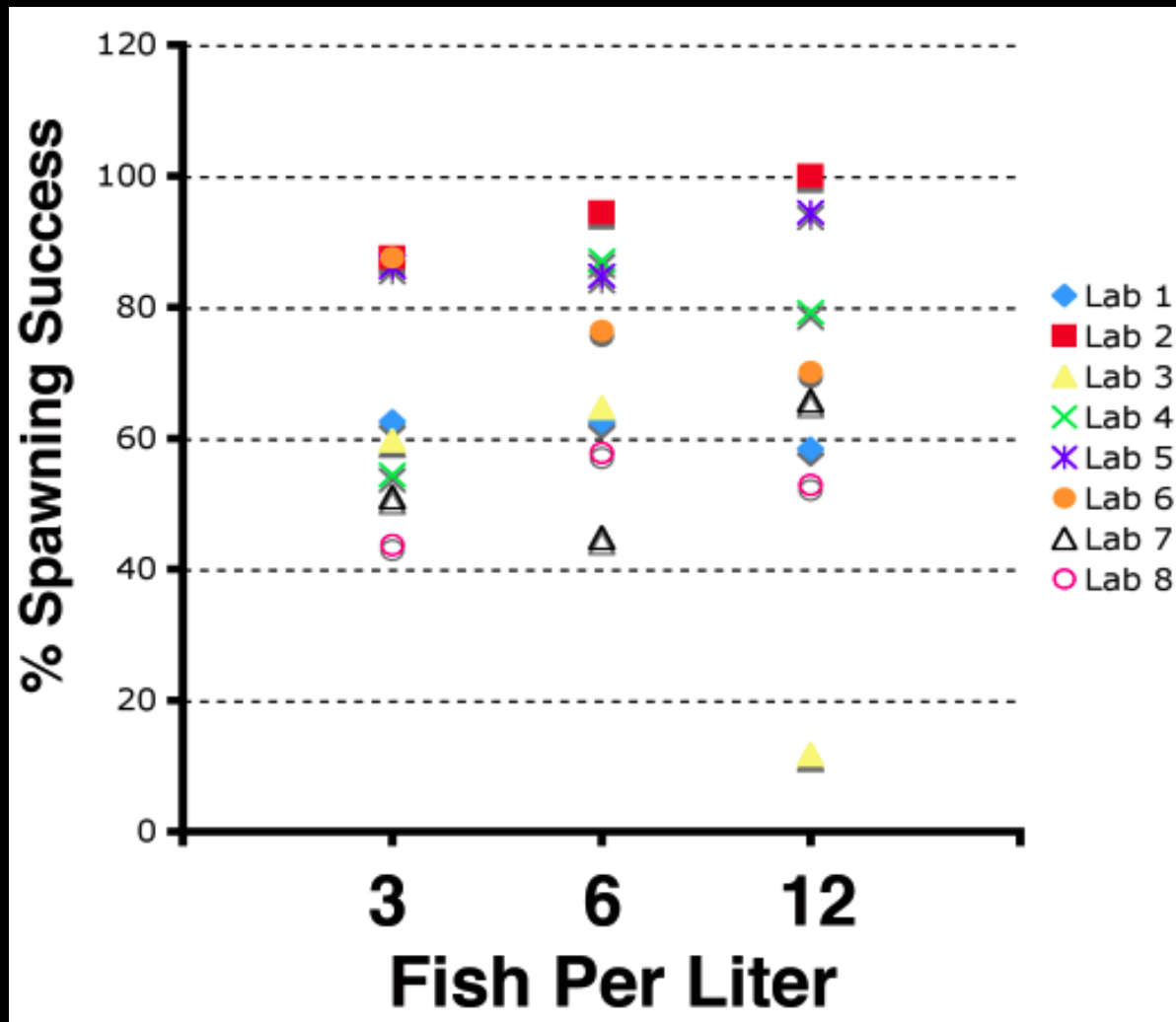
## % viable at 1DPF



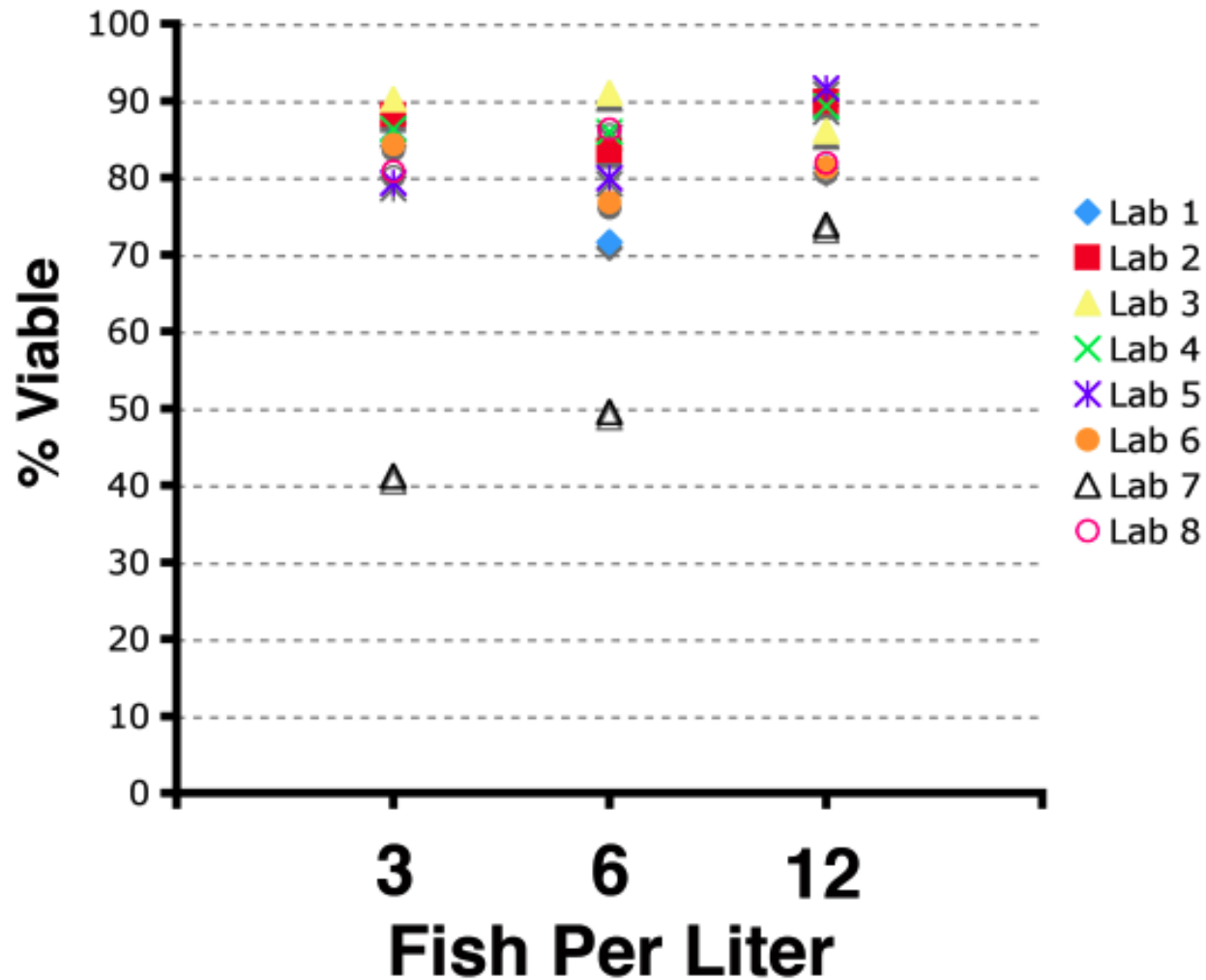
# Average Clutch Size



# Percent Spawning Success



# Percent Viable at 1DPF





# ZHA Density Study Summary

- No differences between treatment densities
- Great variability among laboratories

**Keeping fish at densities as high as 12 fish/l doesn't have  
A negative impact on reproductive performance!**



# Acknowledgements

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## Working Group on Spawning and Reproduction

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# Questions?

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